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Wetland health: big discovery about a tiny organism

photo by LG Patterson

Zhiqiang Hu, assistant professor of civil and environmental engineering, and doctoral student Atreyee Das, have discovered that the presence of a tiny microbe can easily predict wetland health, and also has implications for mitigated wetlands and wastewater treatment.

Microbe plays major role in wetland research

by Jan Wiese-Fales

As organisms go, the microbes that are the heroes of this healthy wetland tale are among the toughest and tiniest. They can live in an environment that lacks both oxygen and sunlight, and roughly four hundred of them will fit into the period at the end of this sentence. University of Missouri Civil Engineering Assistant Professor Zhiqiang Hu and students working in his lab have observed that their presence in wetlands is a novel indicator to effectively and rapidly predict an ecosystem's health. The implication that the introduction of these microbes into wetlands may potentially speed up their successful operation is exciting, and their use in wastewater treatment is promising as well.

Why worry about wetlands?

The term "wetlands" describes a variety of situations from areas that are constantly under standing water to those that have periodically saturated soils. Typically, they are characterized by vegetation that has adapted to these anaerobic soil conditions: cattails, water lilies, reeds, sedges and duckweed. According to the Missouri Department of Natural Resources (MDNR), Missouri has eight types of natural wetlands: swamps, shrub swamps, forested wetlands, marshes, wet meadows, fens and seeps, pond and lake borders and stream banks.

Beyond serving as a critical feature in the overall picture of healthy watersheds and clean water, wetlands are key to plant and animal biodiversity and also have recreational value.

Wetlands play important roles in flood control. They can control erosion and also improve water quality by reducing the levels of soluble pollutants in runoff and overflow.

The Missouri Department of Conservation (MDC) estimates that Missouri's wetlands are the primary habitat for 200 plant and animal species that are considered rare or endangered. Our state's 43 species of amphibians depend on wetlands to complete their life cycles. Additionally, it is estimated that 60 percent of all migratory waterfowl in this country use the Mississippi Flyway corridor on their annual pilgrimages, and are reliant upon Missouri's wetlands for both food and shelter.

This rich diversity makes wetlands popular recreational areas for birdwatchers, nature enthusiasts, hunters and fishermen.

The bad news is that half of all of the country's wetlands have been lost to agricultural, commercial, industrial and residential development; MDC reports that fully 87 percent of Missouri's natural wetlands have disappeared.

Section 404 of the Clean Water Act regulates the impact of development on wetlands, and requires "compensatory mitigation" if adverse impact cannot be avoided or minimized, including wetland restoration, establishment, enhancement or preservation. However, it is not easy to mimic the delicate natural balance of a healthy wetland.

Means and methods

With funding assistance from Region 7 of the U.S. Environmental Protection Agency, MDNR's Water Resource Center partnered with Hu to monitor and assess Missouri wetlands, beginning in the spring of 2008.

MDNR installed instruments in six diverse wetland areas across the state to remotely gather water level and meteorological data for a better understanding of Missouri wetland systems. Two state parks, Pershing and Van Meter, and four conservation areas, Bee Hollow, Four Rivers, Little Bean Marsh and Marion Bottoms, are included in their research observations.

"The department selected the wetland sites because they are some of the best functioning, high value wetlands in Missouri and are located in protective areas," said Renee Bungart, MNDR's deputy communications director. "MU is focusing its efforts on studying the microbiology of the soils during this project and the Department of Natural Resources is focusing its efforts on the hydrologic cycle of the wetlands."

"To some extent, water is always affected by human activity. This is part of our expertise," said Hu of his lab's involvement with the wetlands project. "Many of my projects are related to water quality management and practice."

Hu, Robert Reed, a research associate professor of civil engineering, and graduate assistants from Hu's lab, led by doctoral student Atreyee Das, have concentrated their efforts on three of MDNR's monitored sites: Pershing State Park, a flood plain for Locust Creek, shrub swamp and bottomland forest in Linn County; Bee Hollow Conservation Area, an oxbow com-

plex created by the East Fork of the Little Chariton in Macon County; and Van Meter State Park, a marsh in Saline County. The team visited the sites across the seasons, taking water and soil samples, assisted by MDNR Hydrologist John Horton.

"They have quite a bit of variation," Hu said of the wetlands being studied. "We are specifically looking at biological indicators, ammonia oxidizing microbes — in the soil and wetland water at the sites."

The microbes that are an integral factor in this research play a role in any healthy wetland's nitrogen cycle. Ammonia is a by-product of decomposition that is naturally present in wetlands' hydric — anaerobic and saturated — soils. Ammonia-oxidizing bacteria (AOB) oxidize the ammonia to nitrite, nitrate-oxidizing bacteria (NOB) oxidize the nitrite to nitrate, which can be used by plants for growth. Some of the nitrites are oxidized by denitrifying bacteria and released back into the atmosphere as nitrogen.

Recently, researchers have discovered that ammonia-oxidizing archaea (AOA) also play a key role in the natural oxidation process, working side-by-side with AOB. It is the presence of these microbes in the sampling that give this research project its exciting results.

"For the soil samples, we used a 25-centimeter core push, dividing the samples into upper and lower layers," said Das of the wetland soil sampling, adding that the group took precautions to keep the soil samples and water samples they collected from being contaminated.

A Missouri Department of Natural Resources monitor used to measure the hydrolic cycle at VanMeter State Park.

Back in the lab, Das explained, DNA was isolated from the samples and two molecular biological techniques were used to generate "fragment profiles" of the organisms in the samples. The first, polymerase chain reaction (PCR) was used to amplify the results, and the second, terminal restriction fragment length polymorphism (T-RFLP), separated and detected fragments, which were graphed using laser detection.

"Using these techniques, I can get results in four or five hours," Das said, adding that acquiring results using conventional methods can take up to three weeks.

What the team found surprising about the results was the high level of AOA in the samples — up to three times higher than AOB in the summer months. Furthermore, AOA remained robust even in winter when AOB, which is sensitive to the cold, was detected at a much lower number.

They tested their results by taking samples from Columbia's constructed wetlands with the same result.

"The archaea are instrumental in bringing about nitrification," said Das. "These findings represent the potential to develop a new health indicator of wetland systems with results that are accomplished using a rapid protocol, a scientific method.

"It has worldwide implications for assessing wetlands and for our federal mitigation laws and the building of compensatory wetlands. This is a new method to see what's working and what isn't," she said.

"In the past, when people have tried to do this biological-based assessment, it's been challenging to use these microorganisms to measure the health of wetlands," said Hu. "They are so small; it is hard to quantify them.

"We thought the microbiology work we developed in the lab would be great in the field. We tested it, and it worked," said Hu. "With these tools, we can better construct wastewater systems, as well."

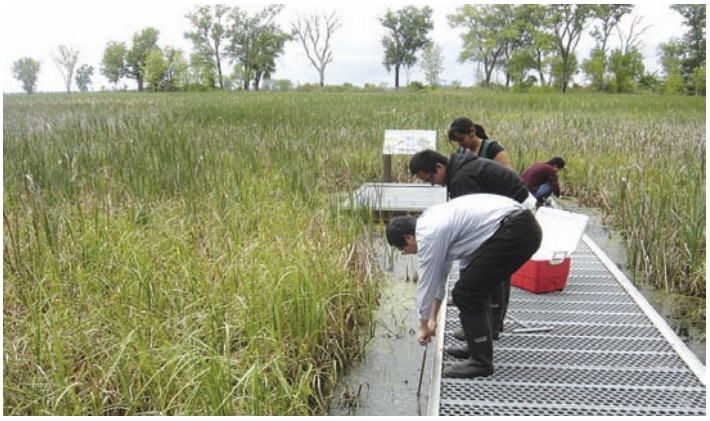
Das said that being able to work with MDNR on the project has been crucial to the research, and that working with Hu has been a rewarding experience. "Dr. Hu has constantly given me ideas on how to improve, and because of this, we've made a lot of progress," said Das, whose educational background is in biology.

"The cool part is that I never thought I could make a big impact in engineering," she said. "But this is an integrated subject. I can use my biology in engineering and the work I am doing will be of use to everyone to make wetlands clean and healthy. Getting an opportunity to make this difference feels good."

"These studies will help the state of Missouri better understand the health and condition of our natural wetlands, which provide habitat for plants and wildlife," said Bungart. "We will use the findings to describe and design future constructed wetlands and will monitor the soils and microbial populations' differences between a constructed wetland and a natural wetland to better learn how to improve water quality through these projects."

Hu is enthusiastic about the potential for their findings for other research, and is considering additional proposals.

"Development of this technology and our results are very relevant to our environment," Hu said, giving the example of eutrophication — the excess release of nutrients into water systems that kills all aquatics. "We will use these results to try to remove nutrients before they enter water bodies. This work is where engineering meets natural environmental systems."



The wetlands research team took soil and water samples from five Missouri wetlands across seasons to assess their function, and in the process made a novel discovery that has far-reaching water quality implications. Below, from left, are doctoral students Atreyee Das and Shashikanth Gajaraj, and Assistant Professor Zhiqiang Hu.

photos courtesy of the Missouri Department of Natural Resources